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In vitro effect of different cleaning procedures of ah plus on microtensile bond strength of immediate or delayed restoration

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ABSTRACT

Aims: Although immediate restoration is recommended after endodontic treatment, delayed restoration is still required in some cases. Also, the presence of the residual endodontic sealer in the pulp chamber may interfere with the adhesive materials and jeopardize the sealability. The purpose of the study was to determine the effect of different sealer residue cleaning methods on dentin bond strength of immediate or delayed endodontic composite restoration.

Methods: The occlusal surface of seven freshly extracted human molar teeth without caries were prepared to expose the flat dentin surface. Teeth were randomly divided into six experimental groups according to the cleaning method and restoration timing. Control group was not contaminated with the endodontic sealer and immediately restored (n=10). Cleaning protocols were cotton pellet soaked in saline (CPS) or cotton pellet soaked in 95% ethanol (CPE) or microbrush soaked in 95% ethanol (MBE). After removing of the sealer, a self-etching adhesive system was applied immediately (i) or delayed (d). The microtensile test applied and data were analyzed by Levene's, two way ANOVA and Tukey HSD statistical tests.

Results: There was no significant difference between the control group and the MBEd and CPSd groups ($p>0.05$). The bond strength of the CPSi, CPEi, CPEd, MBEi groups was statistically different from control ($p<0.05$) and there was no significant difference between these groups ($p>0.05$).

Conclusion: CPSd and the MBEd groups showed better microtensile bond strength values than other experimental groups.

Keywords: Root canal therapy, tensile strength, root canal sealents

INTRODUCTION

A combination of the gutta-percha and root canal sealer is generally used for root canal filling.[1] Root canal sealers are mainly responsible for filling irregularities in the prepared root canal walls as well as gap between gutta-percha and root canal walls.[2,3] Ensuring hermetically obturation prevent the root canal system from bacterial leakage apically or coronally and entomb residual microorganisms.[3,4] Also, residues of endodontic sealer in the pulp chamber can lead to dental discoloration and negatively affect bonding strength of adhesive system to hard tooth surfaces, hence, integrity of coronal seal is impaired, and the prognosis of the root-filled tooth is negatively affected.[5-9]

Adhesive materials generally maintain immediate seal and strengthening of the non-vital tooth, so they are preferred for restoration of the endodontically treated teeth.[7,10,11] So, the aim of this in vitro study was to evaluate influence of the different cleaning protocols on microtensile bond strength for dentin contaminated by the root canal sealers. Among endodontic sealers; epoxy resin based sealers, especially AH Plus (Densply), are generally preferred by clinicians as endodontic sealer[12] and used in various obturation techniques[13] for advantages like antimicrobial property, adequate working time, good sealability and easy handling.[14-16] So, our hypothesis is that after cleaning AH Plus with various methods, bond strength to dentin isn't decrease when immediately or delayed restoration.

METHODS

In this study, 7 intact human molars without cavities, extracted with patient consent during routine dental operations, were used. The study does not contain personal data, the study was conducted only on teeth in the laboratory. Therefore, it does not require ethics committee approval. All procedures were carried out in accordance with the ethical rules and the principles. Experimental groups consisted of three different cleaning methods and their combinations of immediate and delayed restoration timings. Cleaning protocols were cotton pellet soaked in saline (CPS) or cotton pellet soaked in 95% ethanol (CPE), microbrush soaked in 95% ethanol (MBE) and restoration timing protocols were immediately (i) and delayed (d).

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For the experimental groups, AH Plus (Dentsply) was prepared according to manufacturer's instructions and then placed over the exposed dentin as a thin layer. After 10 minutes applied sealer was set then CPS, CPE and MBE removal techniques were applied until the sealer on the dentine surface was unseen. After removing of the sealer, a self-etching adhesive system was applied immediately (i) or delayed (d).

For immediate groups (i) the surface was washed with water spray and air spray dried. A self-etching universal adhesive system (Prime&Bond; Dentsply) was applied to the dentin surface according to the manufacturer's instructions. After curing the adhesive agent with LED curing unit (VALO, Ultradent, Standard mode, 1000 mW/cm^2), resin composite material (Nova Compo B, Imicryl) with 4 mm thickness was placed using incremental technique on the adhesive applied dentin surface and cured with a same LED curing unit (VALO, Ultradent, Standard mode, 1000 mW/cm^2) according to the manufacturer's instructions.

For delayed restoration groups (d), the surface-cleaned teeth were kept in a petri dish containing moist cotton at 30°C for 72 hours. Then, the same restoration steps as in groups (i) were applied.

For the control group, AH Plus endodontic sealer was not applied to the dentin surface and immediate restoration was carried out.

The resin composite placed teeth were sectioned longitudinally, in mesio-distal then bucco-lingual direction using low-speed diamond saw under water cooling (Isomet; Buehler) to obtain beams that have approximate 1 mm^2 cross section area. 10 sticks were obtained from each group. The procedure for obtaining the sticks is summarized in **Figure 1**. The width (w) and length (l) of the beams were measured by electronic caliper (Starret 727-6/150; Starret) prior to the microtensile test to determine the bonding surface area more precisely. The beams are fixed to the microtensile test device (Bisco; Schaumburg) from their ends by cyanoacrylate resin glue, then subjected to a tensile force with speed of 1 mm/min. The force causing the failure was measured and noted in Newtons (N). The microtensile strength value by megapascal (MPa) of beams was calculated with the following formula:

After the microtensile test, specimens were removed from microtensile grip cautiously using a scalpel blade, and the fracture area of each beam was observed using a digital microscope (VHX-900F; Keyence) at × 40 magnification to classification of the fracture mode (adhesive, cohesive or mixed).

The sample size was calculated using G*Power software with 80% power, 0.05 α error probability and 0.4 effect size. Statistical analysis was performed with SPSS (IBM) software. Homogeneity of variances was evaluated with Levene's test, differences between groups were examined with two way ANOVA and post hoc Tukey HSD analyses. Probability value was accepted as 0.05.

RESULTS

A two-way ANOVA was performed to analyze the effect of cleaning protocol and restoration timing on microtensile bond strength. Both parameters have a statistically significant effect on microtensile bond strength and results given in **Table 1** ($p<0.01$). The arithmetic means of the microtensile values obtained from the tested groups was calculated and it was determined that the highest bond strength was in the control group that was not treated with AH plus and did not apply cleaning method. According to statistical analysis, it was determined that there was no significant difference between the control group and the MBEd and CPSd groups ($p>0.05$). The bond strength of the CPSi, CPEi, CPEd, MBEi groups was statistically different from control ($p<0.05$) and there was no significant difference between these groups ($p>0.05$). Box plot test data are shown in **Figure 2**. In the examination with the digital microscope of the separation surfaces of the beams after microtensile test, it was observed that all samples were subjected to adhesive failure.

Table 1. Two way ANOVA results

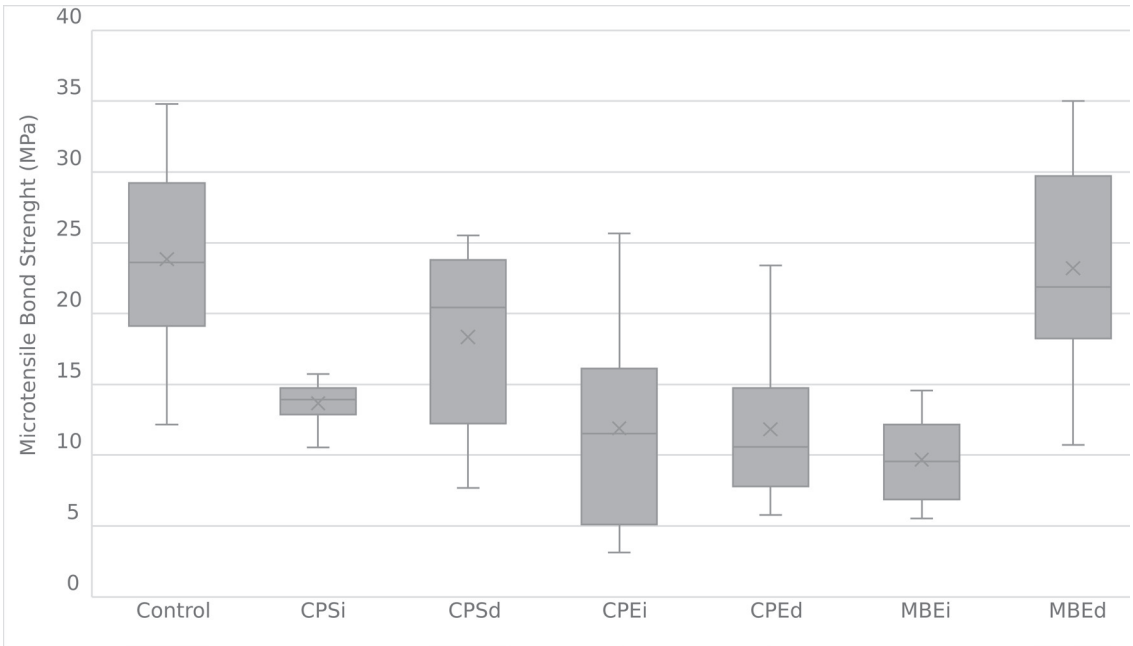

Figure 1. Obtaining sticks for microtensile test.

	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Restoration timing	546.596	1	546.596	15.410	.000	.197
Cleaning protocol	1348.953	3	449.651	12.677	.000	.376
Restoration timing * cleaning protocol	476.386	2	238.193	6.715	.002	.176

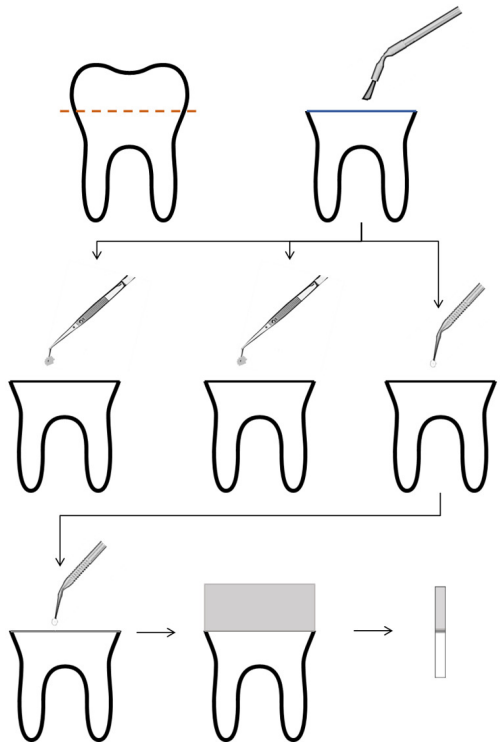

Figure 2. Mean microtensile bond strength of control and experimental groups.

The types of failure modes are presented in **Table 2**. Failure at the the adhesive interface occurred at all of the groups. However, cohesive failure was found only in the CPSi, CPEi, CPEd and MBEi groups.

Table 2. Fracture modes.

	Adhesive (%)	Cohesive (%)	Mix (%)
Control	50.00	0	50
CPSi	46.15	8	46
CPSd	90.00	0	10
CPEi	58.33	17	25
CPEd	87.50	13	0
MBEi	33.33	33	33
MBEd	50.00	0	50

DISCUSSION

In present study, influence of the different root canal sealer residue cleaning protocols on the bond strength to dentin investigated. So, our hypothesis is that after cleaning AH Plus with various methods, bond strength to dentin isn't decrease when immediately or delayed restoration was rejected. In order to test the bond strength, the microtensile method, which was stated to be reliable with previous studies, was chosen[17,18] and molar teeth were preferred because they have a large dentin surface that facilitates stick preparation.

Root canal treated teeth are weakened by caries and subsequent treatment procedures. For this reason, they are recommended to be restored quickly against the risk of fracture and coronal leakage.[19] Coronal leakage is the transport of bacteria, endotoxins, and oral fluids into the canal through the coronal opening as a result of loss of temporary filling material and poorly sealed permanent restorations. It's one of the major causes of endodontic failure.[20] These microleakages cause recurrent caries and apical periodontitis, which jeopardizes the success of both root canal and restorative treatment.[21,22] Clinicians can perform permanent restoration immediately after endodontic treatment or place temporary fillings to wait for confirmation of healing of apical periodontitis.[23,24] With the improvement in the techniques used in endodontic treatment (rubber dam use, magnifying devices, electronic apex locaters and motor-operated rotating files) the success rate of endodontic treatment has increased, and the time required for treatment has been shortened. Thus, single-session root canal treatments became possible. It has also been reported that there is no difference in the success rate and post-operatieve pain in the patient between single session or multi-session endodontic treatment.[25] But in our study, delayed restoration method generally had better dentin bonding strength than immediate restoration.

Clinically, after root canal treatment, the access cavity can be restored using a variety of methods, including direct and indirect restorations. Amalgam restorations, composite resin restorations, coronal-radicular restorations and post applications are conventional treatment methods that can be applied after root canal treatment.[26,27] Also, it has been shown that more harmonic stress distribution is achieved when materials that can be adhered to dental tissues. In addition, it has been reported that if the elasticity module of the restorative material is close to the dental tissues will reduce the stress occur in the remaining tooth tissues due to chewing forces. So, the most ideal restorative material for the restoration to be applied after root canal treatment is appears to be composite resin.[28] After obturation of the root canal system, self-etching adhesives are frequently used for providing a coronal sealing due to relatively easy to application and requiring less technical sensitivity.[29] Nevertheless, there is no chemical bond between epoxy-based resins and methacrylate-based resins and epoxy resin sealers that are not completely removed prior to coronal restoration may cause incomplete polymerization of monomers, bond deterioration, microleakage at the interface and coronal discoloration.[5,30,31] In vitro studies have been conducted with different cleaning methods and materials on the removal of endodontic sealers. The most studied model was dry cotton pellets and organic solvents. They are followed by mechanical drill refinement, air abrasion, microabrasion, ultrasound. It has been found that after cleaning using dry cotton pellets, the bond strength decreased to 69-88% of uncontaminated controls.[1,12,32-34] Alongside with numerous organic solvents, formamide provides high cleaning efficacy, but its potential teratogenicity has been noted in studies. Another solvent, acetone, affects the bond strength in a negative way because of collagen fiber collase.[35] In clinical practice, ethanol and distilled water is a routinely used solvent for surface cleaning

due to its easy accessibility. In present in-vitro study, the highest bonding was observed in the delayed-ethanol micropellet and delayed- wet cotton group (**Figure 2**). When using cotton pellets soaked with ethanol, it was observed that the immediate restoration provided a better bonding strength. Cleaning AH Plus contaminated dentin with ethanol-saturated cotton pellets appears to be ineffective for creating coronal seal with self-etching adhesives. In present study, the lowest adhesion was observed in the group in which ethanol was used with micropellet and immediately adhesive was applied. However, the highest bonding values were found with micropellet using ethanol and applying adhesive after 72 hours. The other group with high adhesion was the group that used distilled water-soaked cotton and applied adhesive after 72 hours. The reason for this is that maybe the after cleaning the Ah-Plus residues, the remaining residues completely set in the dentinal tubules and do not interfere the bonding agent during adhesive procedure.

As a limitation, effects of the presence of saliva in the oral environment, variable temperatures, mechanical fatigue, etc. were not evaluated due to lack of time and resources. In future randomized clinical trials of this subject may be beneficial.

CONCLUSION

Within the limitation of the present study, CPSd and the MBEd groups showed better bond strength values than other experimental groups. After root canal treatment, the bonding strength of the adhesive restoration to dentin can be increased with use of the effective sealer cleaning method and the application of adhesive restoration at the appropriate time.

ETHICAL DECLARATIONS

Ethic Committee Approval: In this study, 7 intact human molars without cavities, extracted with patient consent during routine dental operations, were used. The study does not contain personal data, the study was conducted only on teeth in the laboratory. Therefore, it does not require ethics committee approval.

Informed Consent: Since this study was conducted in the laboratory on 7 intact human molars without decay, extracted with patient consent during routine dental operations, it does not require informed consent.

Referee Evaluation Process: Externally peer-reviewed.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

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